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#13

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

RE: U.S. Patent No. 4,562,073

ISSUED: December 31, 1985

Box Pat. Ext.

TO: Ronald G. MICETICH et al.

FOR: PENICILLIN DERIVATIVES

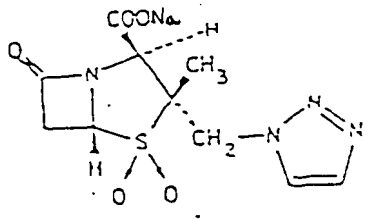
Honorable Commissioner of Patents and Trademarks
Washington, D.C. 20231

APPLICATION FOR EXTENSION OF PATENT TERM
UNDER 35 U.S.C. 156 AND 37 C.F.R. 1.710

Pursuant to Section 201(a) of the Drug Price Competition and Patent Term Restoration Act of 1984, 35 U.S.C. Sec 156, Taiho Pharmaceutical Company Ltd., owner of the above identified patent, hereby requests a 1358 day extension of the patent term of United States Patent No. 4,562,073, covering ZOSYN®, Sodium [2S-(2α,3β,5α)]-3-methyl-7-oxo-3-(1H-1,2,3-triazol-1-ylmethyl)-4-thia-1-azabicyclo [3.2.0] heptane-2-carboxylate 4,4-dioxide (CAN) granted to Ronald G. Micetich, Shigeru Yamabe, Motoaki Tanaka, Makoto Kajitani, Tomio Yamazaki and Naobumi Ishida, on December 31, 1985. Taiho Pharmaceutical Company Ltd.'s ownership of the patent is evidenced by an assignment recorded in the U.S. Patent Office at Reel 4161, Frame 0964. Lederle Laboratories, which applied for the commercial marketing approval, is the licensee of the applicant under the patent in the United States of America.

Applicant submits this application for the extension of the patent term of U.S. Patent No. 4,562,073 by providing the following information organized corresponding to 37 CFR 1.740.

(1) The approved product is identified as ZOSYN®. ZOSYN® contains, as an active ingredient, tazobactam sodium, whose chemical name is Sodium [2S- (2α,3β,5α)] -3-methyl-7-oxo-3-(1H-1,2,3-triazol-1-ylmethyl)-4-thia-1-azabicyclo[3.2.0] heptane-2-carboxylate 4,4-dioxide (CAN) which is a compound having the following structural formula:



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(2) The regulatory review period occurred under section 507 of the Federal Food, Drug and Cosmetic Act (FFDCA) (21 USC §357). Section 507 provides for the submission and approval of new drug applications (NDAs) for antibiotic drugs.

(3) ZOSYN® was approved by the Food and Drug Administration (FDA) for commercial marketing under section 507 of the FFDCA on October 22, 1993.

(4) ZOSYN® contains, as an active ingredient, tazobactam sodium. This active ingredient has not previously been approved for commercial marketing or use under 507 of the FFDCA. The other active ingredient in ZOSYN® is piperacillin sodium which had been previously approved for commercial marketing on December 29, 1981 (NDA 50-545) for use in treatment of bacterial infections under Section 507 of the Federal Food, Drug and Cosmetic Act (21 U.S.C. §357).

(5) The product was approved for commercial marketing on October 22, 1993. This application is being submitted within the permitted period, the last day within the sixty day period permitted for submission of an application for extension is December 20, 1993.

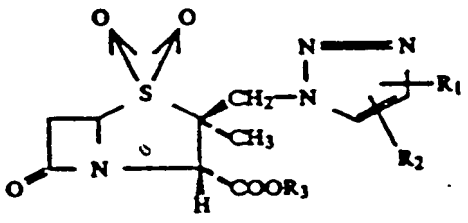
(6) The patent for which patent term extension is sought is U.S. Patent No. 4,562,073, which issued on December 31, 1985, naming Ronald G. Micetich, Shigeru Yamabe, Motoaki Tanaka, Makoto Kajitani, Tomio Yamazaki and Naobumi Ishida as the inventors for PENICILLIN DERIVATIVES. The term of the patent has never been extended and has not yet expired. The unextended patent will expire on July 16, 2002.

(7) A complete copy of U.S. Patent No.4,562,073 in the prescribed form is attached as Attachment 1.

(8) A copy of the Terminal Disclaimer disclaiming the portion of the patent subsequent to July 16, 2002 is attached as Attachment 2. A copy of the maintenance fee statements for U.S.Patent 4,562,073 indicating that the maintenance fees have been paid is attached as Attachment 3.

(9) U.S. Patent No. 4,562,073 claims the approved product. Specifically, the active ingredient tazobactam is claimed in claim 1, which follows:

> 1. A penicillin derivative represented by the following formula



wherein R_1 is hydrogen or trialkylsily; R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1, 3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt; and R_3 has the same meaning as above R_2'.

Tazobactam is covered in dependent claim 8 as follows:

8. The penicillin derivative as defined in claim 1 wherein R_3 is a group for forming a pharmaceutically acceptable salt.

Tazobactam is covered in dependent claim 10 as follows:

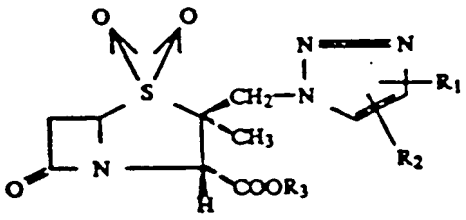
10. The penicillin derivative as defined in claim 8 wherein the group for forming the pharmaceutically acceptable salt represented by R_3 is alkali metal atom, alkaline earth metal atom or ammonium, or the group $COOR_3$ represents a carboxylic acid salt formed from the carboxyl group and a member selected from the group consisting of cyclohexylamine,trimethylamine, diethanolamine, arginine and lysine.

Tazobactam is also covered in dependent claim 11 as follows:

11. The penicillin derivative as defined in claim 1 wherein R_1 and R_2 are hydrogen.

and by dependent claims 17 and 18 which further recite a "group for forming a pharmaceutically acceptable salt" for R_3. ZOSYN®, as a pharmaceutical composition containing a combination of tazobactam sodium and piperacillin sodium, a β-lactam antibiotic, is also covered by claim 15 as follows:

15. A pharmaceutical composition useful for treating bacterial infections in mammals, said composition comprising (A) a β-lactam antibiotic and (B) a compound of the formula

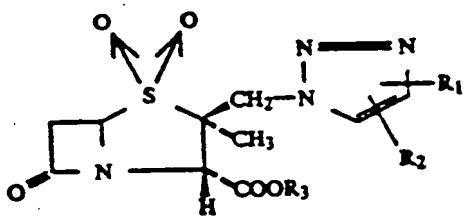


wherein R_1 is hydrogen or trialkylsilyl; R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl) carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt; and R3 has the same meaning as above R_2', the weight ratio of (A)/(B) being 0.1 to 10, said β-lactam antibiotics being selected from the group consisting of ampicillin, amoxicillin, hetacillin, ciclacillin, mecillinam, carbenicillin, sulbenicillin, ticarcillin, piperacillin, apalcillin, methicillin, mezlocillin, bacampicillin, carindacillin, talampicillin, carfecillin and pivmecillinam, cephaloridine, cephalothin, cephapirin, cephacetrile, cefazolin, cephalexin, cefradine, cefotiam, methicillin, mezlocillin, bacampicillin, carindacillin, talampicillin, carfecillin and pivmecillinam, cephaloridine, cephalothin, cephapirin, cephacetrile, cefazolin, cephalexin, cefradine, cefamandole, cefuroxime, cefoxitin, cefmetazole, cefsulodin, cefoperazone, cefotaxime, ceftizoxime, cefmenoxime, latamoxef, cefaclor, cefroxadine, cefatrizine, cefadroxil and cephaloglycin; and pharmaceutically acceptable salts thereof.

(ZOSYN® contains a combination of piperacillin sodium and tazobactam sodium wherein the weight ratio of piperacillin/tazobactam is 8:1, which is within the range of 0.1 to 10 as recited in the claim).

ZOSYN® is approved for the treatment of certain bacterial infections. This use is covered by claim 16 which recites the method of treating bacterial infections by administering a pharmaceutical composition containing tazobactam and a β-lactam antibiotic as follows:

16. A method of treating a bacterial infection in a mammal subject, said method comprising administering to said subject (A) a β-lactam antibiotic and (B) a compound of the formula



wherein R_1 is hydrogen or trialkylsilyl; R_2 is hydrogen, trialkylsilyl or COOR'$_2$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or un-substituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt; and R_3 has the same meaning as above R_2', the weight ratio of (A)/(B) administered being 0. 1 to 10, said β-lactam antibiotics being selected from the group consisting of ampicillin, amoxicillin, hetacillin, ciclacillin, mecillinam, carbenicillin, sulbenicillin, ticarcillin, piperacillin, apalcillin, methicillin, mezlocillin, bacampicillin, carindacillin, talampicillin, carfecillin and pivmecillinam, cephaloridine, cephalothin, cephapirin, cephacetrile, cefazolin, cephalexin, cefradine, cefotiam, cefamandole, cefuroxime, cefoxitin, cefmetazole, cefsulodin, cefoperazone, cefotaxime, ceftizoxime, cefmenoxime, latamoxef, cefaclor, cefroxadine, cefatrizine, cefadroxil and cephaloglycin; and pharmaceutically acceptable salts thereof.

(10) The relevant dates and information pursuant to 35 USC 156(g) are as follows:

(a) IND No. 31,705 submitted to FDA on June 10, 1988
(b) Effective date of IND No. 31,705 = July 10, 1988
(c) NDA No. 50-684 submitted to FDA on August 30, 1991
(d) NDA No. 50-684 approved by FDA on October 22, 1993

(11) A brief description of the significant activities undertaken by the applicant's licensee during the applicable regulatory review period with respect to the approved product and the significant dates applicable to such activities is attached as attachment 4.

(12) In the opinion of the applicant, this patent is eligible for the requested 1358 day extension because, the term of the patent has not yet expired, the term of the patent has never been extended, the application for extension is submitted by the owner of record of the patent, the product has been subject to a regulatory review period before its commercial marketing or use, and permission for the commercial marketing of the product is the first permitted commercial marketing of the product under the provision of law under which the regulatory review occurred.

The regulatory review period started on July 10, 1988, the day that IND 31,705 became effective under section 507(d) of the FFDCA. This was subsequent to the issuance of Patent No. 4,562,073 on December 31, 1985.

(i) The regulatory review period under 35 U.S.C. §156 (g)(1)(B) began July 10, 1988 and ended October 22, 1993, which is a total of 1931 days, which is the sum of (ii) and (iii) below:

(ii) The period of review under 35 U.S.C. §156(g)(1)(B) (i), the "Testing Period", began on July 10, 1988 (since the IND was filed 30 days prior to test date and was not disapproved by the FDA during that period), and ended on August 30, 1991, which is 1147 days (half this period is 573.5).

(iii) The period of review under 35 U.S.C. §156(g)(1)(B) (ii), the "Application Period", began on August 30, 1991, and ended October 22, 1993, which is 784 days.

The regulatory review period upon which the period of extension is calculated is the entire regulatory review period, as determined above (1931 days) less:

(i) the number of days in the regulatory review period which were on or before the date on which the patent issued, December 31, 1985, which is zero (0) days; and

(ii) the number of days during which applicant did not act with due diligence, which is zero (0) days; and

(iii) one-half the number of days determined in subparagraph 12(ii), the "Testing Period" above after subtracting therefrom the number of days which were on or before the date on which the patent issued and the number of days during which applicant did not act with due diligence (zero in total) or 573 days.

1931 - 573 = 1358 days

The number of days as determined above (1358 days) when added to the original term of the patent would result in the new patent expiration date of April 3, 2006.

Fourteen (14) years, when added to the date of NDA approval (October 22, 1993), would result in the date of October 22, 2007.

The issuance of the original patent and the submission of the request for exemption occurred after September 24, 1984. Thus the period of the limitation under 35 U.S.C. §156(g)(6)(A) is five (5) years. Five years, when added to the original expiration date of the patent (July 16, 2002), would result in the date July 16, 2007.

The earlier date, as determined above, is April 3, 2006.

Therefore, the length of extension of the patent term claimed by applicant is 1358 days.

(13) Applicant acknowledges the duty to disclose to the commissioner of Patents and Trademarks and the Secretary of Health and Human services any information which is material to the determination of entitlement to the extension sought in this application for extension of the patent term.

(14) The fee of $1000.00 is enclosed with this application.

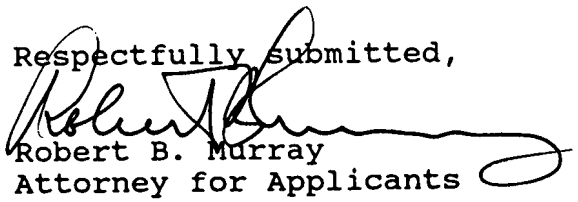
(15) Inquiries and correspondence relating to this application for extension are to be directed to:

Robert B. Murray
Nikaido, Marmelstein, Murray & Oram
655 Fifteenth St. N.W. Suite 330
Washington, D.C. 20005-5701
(202) 638-5000

(16) This application for extension of the patent term is being submitted in duplicate, as certified below.

(17) The undersigned hereby declares that he is a patent attorney authorized to practice before the United States Patent and Trademark Office and has general authority from applicant, Taiho Pharmaceutical Company, Limited (power of attorney attached as Attachment 5), for the purpose of transacting all matters reasonably related to obtaining an extension of the patent term for U.S. Patent No. 4,562,073, to act on its behalf in patent matters; that he has reviewed and understands the contents of the application being submitted pursuant to 35 USC 156; that he believes the patent is subject to extension pursuant to 37 CFR 1.710; that he believes an extension of the length claimed is fully justified under 35 USC 156, and the applicable regulations; and that he believes the patent for which the extension is being sought meets the conditions for extension of the term of a patent as set forth in 37 CFR 1.720.

Respectfully submitted,



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ATTACHMENT 1

United States Patent [19]

Micetich et al.

[11] Patent Number: 4,562,073

[45] Date of Patent: * Dec. 31, 1985

[54] PENICILLIN DERIVATIVES

[75] Inventors: Ronald G. Micetich, Alberta, Canada; Shigeru Yamabe, Kobe, Japan; Motoaki Tanaka, Tokushima, Japan; Makoto Kajitani, Tokushima, Japan; Tomio Yamazaki, Tokushima, Japan; Naobumi Ishida, Tokushima, Japan

[73] Assignee: Taiho Pharmaceutical Company Limited, Tokyo, Japan

[*] Notice: The portion of the term of this patent subsequent to Jul. 16, 2002 has been disclaimed.

[21] Appl. No.: 519,491

[22] Filed: Aug. 1, 1983

[30] Foreign Application Priority Data

Dec. 24, 1982 [JP] Japan 57-233967
Feb. 10, 1983 [JP] Japan 58-21200

[51] Int. Cl.⁴ C07D 499/00; A61K 31/425
[52] U.S. Cl. ..... 424/114; 260/245.2 R; 514/192
[58] Field of Search ..... 260/245.2 R, 245.2 T; 424/270, 271, 114; 514/192

[56] References Cited

U.S. PATENT DOCUMENTS

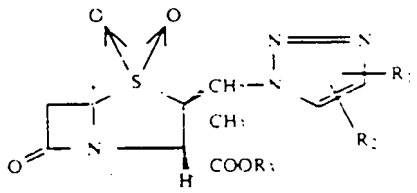
4,331,677 5/1982 Foglio et al. ..... 260/245.2 R

Primary Examiner—Nicholas S. Rizzo

Attorney, Agent, or Firm—Murray, Whisenhunt and Ferguson

[57] **ABSTRACT**

A penicillin derivative represented by the following formula



wherein R_1 is hydrogen or trialkylsilyl; R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-15} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt; and R_3 has the same meaning as above R_2'.

18 Claims, No Drawings

PENICILLIN DERIVATIVES

This invention relates to penicillin derivatives and to a process for preparing them.

Of the commercially available antibiotics, β-lactam type antibiotics having a β-lactam ring, namely penicillins and cephalosporins, are best known and frequently used. Although widely used as useful chemotherapeutic drugs, the β-lactam type antibiotics can not achieve satisfactory effects against some types of microorganisms because of resistance of the microorganism to the β-lactam type antibiotics. The resistance thereof are usually attributable to β-lactamase produced by the microorganism. The β-lactamase is an enzyme which acts to cleave the β-lactam ring of the β-lactam type antibiotic, thereby causing the antibiotic to lose its antimicrobial activity. For this reason, the action of β-lactamase must be eliminated or inhibited so as to enable the β-lactam type antibiotic to produce satisfactory effects. The elimination or inhibition of the β-lactamase activity can be achieved by β-lactamase inhibitors, which are used conjointly with the β-lactam type antibiotic to increase the antimicrobial activity of the antibiotic.

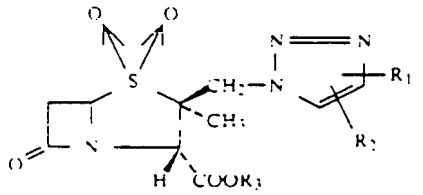
It is an object of the present invention to provide novel compounds having β-lactamase inhibitory action.

It is another object of the invention to provide processes for preparing the same.

It is a further object of the invention to provide a pharmaceutical composition having excellent β-lactamase inhibitory action.

It is an additional object of the invention to provide compositions which, when combined with β-lactam type antibiotics, can increase the antibacterial activity of the antibiotics.

The penicillin derivatives of the present invention are represented by the formula



(I)

wherein R_1 is hydrogen or trialkylsilyl, R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl and group for forming a pharmaceutically acceptable salt; and R_3 has the same meaning as R_2'.

The penicillin derivatives of the present invention are all novel compounds and have β-lactamase inhibitory properties, hence useful as β-lactamase inhibitory agents.

The penicillin derivatives of the invention, when used in combination with a known β-lactam type antibiotic,

can increase the antimicrobial activity of the β-lactam type antibiotic.

Examples of antibiotics which can be used conjointly with the compounds of the present invention are β-lactam antibiotics which exhibit antibacterial action against gram-positive or gram-negative bacteria and which include commonly used penicillins such as ampicillin, amoxicillin, hetacillin, ciclacillin, mecillinam, carbenicillin, sulbenicillin, ticarcillin, piperacillin, apalcillin, methicillin, mezlocillin and salts thereof; esters of penicillins such as bacampicillin, carindacillin, talampicillin, carfecillin and pivmecillinam; cephalosporins such as cephaloridine, cephalothin, cephapirin, cephacetrile, cefazolin, cephalexin, cefradine, cefotiam, cefamandole, cefuroxime, cefoxitin, cefmetazole, cefsulodin, cefoperazone, cefotaxime, ceftizoxime, cefmenoxime, latamoxef, cefaclor, cefroxadine, cefatrizine, cefadroxil, cephaloglycin, and salts thereof. The β-lactam antibiotics are usually used in an amount of about 0.1 to about 10 parts by weight, preferably about 0.2 to about 5 parts by weight, per part by weight of the compound of the invention.

Examples of the trialkylsilyl groups represented by R_1 and R_2 in the formula (I) include trialkylsilyl having straight-chain or branched-chain C_{1-6} alkyl such as methyl, ethyl, propyl, isopropyl, butyl, tert-butyl, pentyl, hexyl, and the like.

Examples of the group R_2' of $COOR_2'$ represented by R_2 in the formula (I) include: C_{1-18} alkyl such as methyl, ethyl, propyl, isopropyl, tert-butyl, pentyl, hexyl, decyl, undecyl, dodecyl, tetradecyl, hexadecyl, octadecyl and like straight- or branched-chain alkyl; C_{2-7} alkoxymethyl such as methoxymethyl, ethoxymethyl, propyloxymethyl, isopropyloxymethyl, butoxymethyl and hexyloxymethyl; C_{3-8} alkylcarbonyloxymethyl such as methylcarbonyloxymethyl, ethylcarbonyloxymethyl, butylcarbonyloxymethyl and hexylcarbonyloxymethyl; C_{4-9} alkylcarbonyloxyethyl such as methylcarbonyloxyethyl, ethylcarbonyloxyethyl, butylcarbonyloxyethyl and pivaloyloxyethyl; (C_{5-7} cycloalkyl)carbonyloxymethyl such as cyclopentylcarbonyloxymethyl, cyclohexylcarbonyloxymethyl and cycloheptylcarbonyloxymethyl; C_{9-14} benzylcarbonyloxyalkyl such as benzylcarbonyloxymethyl, benzylcarbonyloxyethyl, benzylcarbonyloxypropyl and benzylcarbonyloxybutyl; C_{3-8} alkoxycarbonylmethyl such as methoxycarbonylmethyl, ethoxycarbonylmethyl, propyloxycarbonylmethyl and hexyloxycarbonylmethyl; C_{4-9} alkoxycarbonylethyl such as methoxycarbonylethyl, ethoxycarbonylethyl, propyloxycarbonylethyl, butoxycarbonylethyl and hexyloxycarbonylethyl; halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms such as chloromethyl, 2,2-dibromoethyl and trichloroethyl; C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl such as p-methoxybenzyl, p-ethoxybenzyl, o-nitrobenzyl and p-nitrobenzyl; (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl such as (2-oxo-1,3-dioxoden-4-yl)methyl, (5-methyl-2-oxo-1,3-dioxoden-4-yl)methyl and (5-phenyl-2-oxo-1,3-dioxoden-4-yl)methyl; C_{8-13} benzoyloxyalkyl such as benzoyloxymethyl, benzoyloxyethyl, benzoyloxypropyl and benzoyloxybutyl; etc.

Examples of the groups represented by R_3 in the formula (I) are the same as those exemplified in respect of the group R_2'.

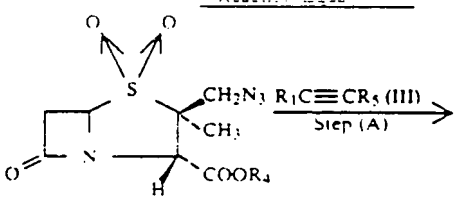
The ester residues represented by R_2' and R_3 include both carboxyl-protecting groups acceptable in the synthesis of penicillin compounds and pharmaceutically

acceptable ester residues. A pharmaceutically acceptable ester having such residue is an ester which is easily hydrolyzed in vivo and which is a non-poisonous ester capable of rapidly decomposing in the blood or tissue of humans, thereby producing the corresponding acid of the formula (I) in which R_3 is hydrogen atom. Generally in the synthesis of penicillin compounds, ester-protecting groups are used in the art to protect penicillin carboxyl groups or other carboxyl groups. While it is difficult to determine which ester-protecting group should be used, consideration are usually given to select esters in which the protecting group per se is sufficiently stable in the reaction and which does not permit cleavage of the β-lactam ring in removal of the ester-protecting groups. Most commonly used as such ester-protecting groups are p-nitrobenzyl group, benzhydryl group, trichloroethyl group, trichlorosilyl group, tetrahydropyranyl group, etc. Examples of the pharmaceutically acceptable ester groups are phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, (2-oxo-1,3-dioxoden-4-yl)methyl, etc.

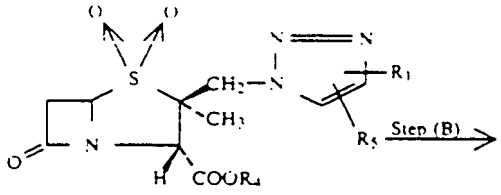
Examples of the group for forming a pharmaceutically acceptable salt represented by R_2' and R_3 in the formula (I) include; sodium, potassium, lithium, or like alkali metal atoms; calcium, magnesium or like alkaline earth metal atoms; cyclohexylamine, trimethylamine, diethanolamine or like organic amine; arginine, lysine or like basic amino acid residues; ammonium residues, etc.

The penicillin derivatives of the present invention having the formula (I) can be prepared by the processes as shown in reaction equations given below. The processes differ according to the kind of the groups represented by R_1 and R_2.

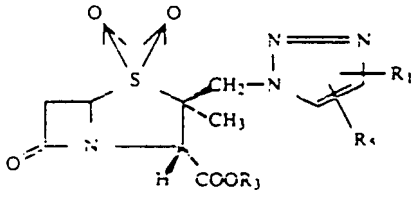
Reaction Equation-1



(II)



(IV)



(I-a)

In the foregoing formulae, R_1 and R_3 are as defined above, R_4 is penicillin carboxyl-protecting group and R_5 is trialkylsilyl or $COOR_2'$ wherein R_2' is as defined above.

Examples of the penicillin carboxyl protecting group expressed by R_4 include known groups such as those

described in Japanese Unexamined Patent Publication No. 81380/1974 and H. E. Flynn, "Cephalosporins and Penicillins, Chemistry and Biology" (published in 1972 by Academic Press). Specific examples thereof are ethyl, propyl, tert-butyl, trichloroethyl and like substituted or unsubstituted alkyl groups; benzyl, diphenyl methyl, p-nitrobenzyl and like substituted or unsubstituted aralkyl groups; acetoxymethyl, acetoxyethyl, propionyloxyethyl, pivaloyloxyethyl, pivaloyloxypropyl, benzyloxymethyl, benzyloxyethyl, benzylcarbonyloxymethyl, cyclohexylcarbonyloxymethyl and like acyloxyalkyl groups, methoxymethyl, ethoxymethyl, benzyloxymethyl and like alkoxyalkyl groups; and other groups such as tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl and like groups.

The steps (A) and (B) of the foregoing process will be described below in detail.

Step (A)

A penicillanic acid derivative of the formula (II) is reacted with an acetylene derivative of the formula (III) to provide a compound of the formula (IV). The reaction is conducted in a suitable solvent by reacting a known penicillanic acid derivative of the formula (II) with a known acetylene derivative of the formula (III) in an amount of about 1 to about 50 moles, preferably about 1 to about 10 moles, per mole of the derivative of the formula (II).

The solvents useful in the reaction are not particularly limited and include any of those which do not adversely affect the reaction. Specific examples of the solvents are an acetylene derivative of the formula (III) as used in excess amount or benzene, toluene, xylene and like aromatic hydrocarbons, tetrahydrofuran, dioxane or like ethers, acetone and like polar organic solvents; etc. These solvents are used singly or in mixture. The reaction proceeds usually at a temperature of between about 50° C. and a boiling point of the solvent, or at a temperature of less than 200° C. in a sealed reactor, and goes to completion in about 2 to about 72 hours.

Depending upon the kind of the penicillin carboxyl protecting group represented by R_4, the compounds of the formula (IV) obtained in step (A) may be esters of the penicillin derivatives of the present invention having the formula (I). The compounds of the formula (IV) are preferably subjected to de-esterification to form a derivative of the formula (I-a) in which R_3 is hydrogen which, in turn, is converted into a pharmaceutically acceptable salt or ester thereof as in the following step (B). The compound of the formula (IV) can also be made into an ester of the formula (I-a) by the conventional ester interchange reaction in the step (B).

Step (B)

The compound of the formula (IV) is subjected to de-esterification without or after isolation from the reaction mixture obtained in step (A), whereby a penicillin derivative of the formula (I-a) in which R_3 is hydrogen is obtained.

As the de-esterification method, reduction, hydrolysis, treatment with an acid and like method can be employed for converting the carboxyl-protecting group to carboxyl group. For example, if the carboxyl-protecting group is an active ester, the reaction frequently proceeds with ease under mild hydrolysis conditions or by merely bringing the ester into contact with water. The reduction method is employed when the carboxyl-

protecting group is trichloroethylbenzyl, p-nitrobenzyl, diphenylmethyl or the like. Treatment with an acid is adopted when the carboxyl-protecting group is 4-methoxybenzyl, tert-butyl, trityl, diphenylmethyl, methoxymethyl, tetrahydropyranyl or the like.

The reduction can be conducted by treating the ester of the formula (IV) with a mixture of (a) zinc, zinc-amalgam or like metal and/or chromium chloride, chromium acetate or like chromium salt and (b) formic acid, acetic acid or like acid. Alternatively, the reduction can be conducted with use of a catalyst in hydrogen atomosphere in a solvent. Examples of the catalysts are platinum, platinum oxide, palladium, palladium oxide, palladium-barium sulfate, palladium-calcium carbonate, palladium-carbon, nickel oxide, Raney-nickel, etc. The solvents are not particularly limited so far as they do not adversely affect the reaction, and include methanol, ethanol and like alcohols; tetrahydrofuran, dioxane and like ethers; ethyl acetate and like esters; acetic acid and like fatty acids; and a mixture of these organic solvents and water.

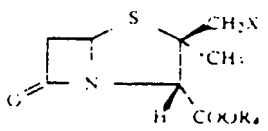
The acids useful for eliminating the carboxyl-protecting group of the ester of the formula (I-a) are formic acid, acetic acid and like lower fatty acids; trichloroacetic acid, trifluoroacetic acid and like trihalogenated acetic acids; hydrochloric acid, hydrofluoric acid and like hydrohalogenic acids; p-toluene-sulfonic acid, trifluoromethane-sulfonic acid and like organic sulfonic acids; and a mixture of these. In this reaction, when the acid used is in a liquid state and acts also as a solvent, it is not necessary to use other solvents. However, dimethylformamide, dichloromethane, chloroform, tetrahydrofuran, acetone and like solvents which do not adversely affect the reaction may be used.

The penicillin derivative of the present invention having the formula (I-a) in which R_3 is hydrogen can be transformed by the salt-forming reaction or esterification commonly employed in the art into a pharmaceutically acceptable salt or ester as contemplated.

If the ester residue is, for example, 3-phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl or like group, the penicillin derivative of the formula (IV) can be alkylated by using 3-halogenated phthalide, 4-halogenated crotonolactone, 4-halogenated-γ-butyrolactone or the like. Suitable halogens of the foregoing halides include chlorine, bromine, iodine, etc. The reaction is carried out by dissolving the salt of the penicillin derivative of the formula (IV) in N,N-dimethylformamide or like suitable polar organic solvent and adding an approximately equimolecular amount of a halide to the solution. The reaction temperature ranges from about 0° to about 100° C., preferably from about 15° to about 35° C. Suitable salts of the penicillin derivative to be used in the esterification are salts of sodium, potassium or like alkali metals; salts of triethylamine, ethyldiisopropylamine, N-ethylpiperidine, N,N-dimethylaniline, N-methylmorpholine or like tertiary amines, etc. After completion of the reaction, the contemplated product can be easily separated by the conventional method and also can be purified, when required, by recrystallization, thin layer chromatography, column chromatography or like method.

The compound of the formula (II) to be used as the starting material in the step (A) is a novel compound undisclosed in literture and can be synthesized by the method described in Japanese Patent Application No. 69142/1982 (relating to an invention accomplished by us). The disclosed method comprises the steps of react-

ing a metal azide with a known derivative of penicillanic acid of the formula



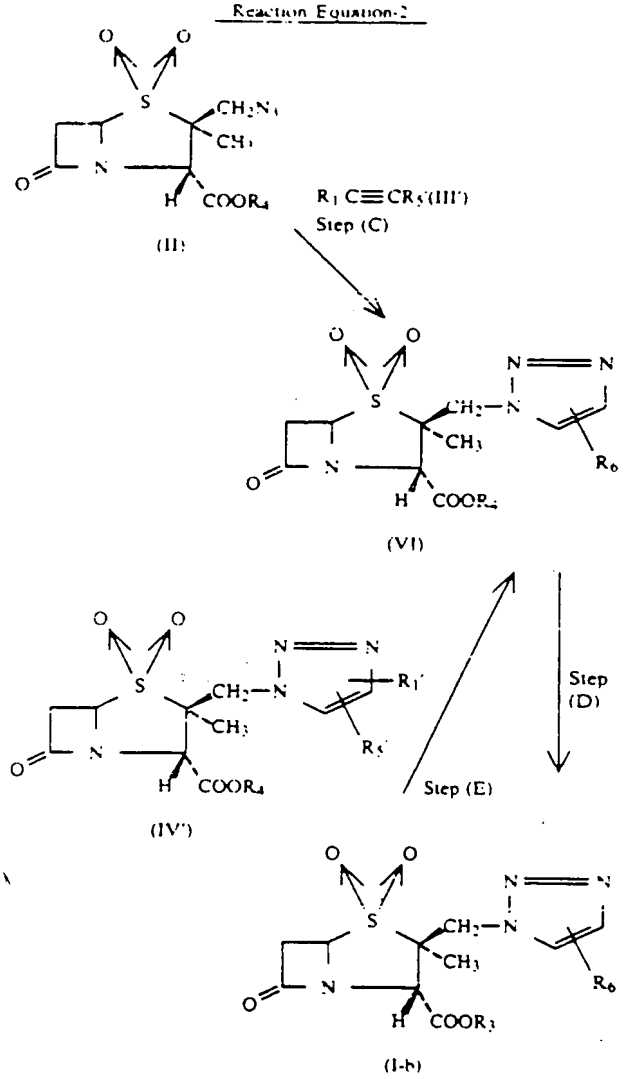
(V)

wherein X represents chlorine atom or bromine atom and R_4 is as defined above, oxydizing the reaction mixture and subjecting the resulting compound to de-esterification.

The foregoing method will be described below in detail. The reaction between the compound of the formula (V) and the metal azide is conducted in a suitable solvent by using the metal azide in an amount of about 1 to about 50 moles, preferably about 1 to about 10 moles, per mole of the compound of the formula (V). Examples of the metal azides which can be used include those commonly used, such as sodium azide, potassium azide and like azides of alkali metals, and barium azide and like azides of alkaline earth metals. Useful solvents are not particularly limited as far as they do not adversely affect the reaction. Examples of useful solvents are dimethylformamide, ethyl acetate, acetone, dichloromethane, tetrahydrofuran, dioxane, methanol, ethanol and like organic solvents. These organic solvents can be used singly or in mixtures. Also a mixture of such solvent and water is usable. The reaction proceeds at a temperature of usually about −20° to about 100° C., preferably about 0° to about 100° C. The resulting product can be used in subsequent oxidation without isolation, or alternatively after isolation and purification by a conventional method. The oxidation subsequent to the azide-forming reaction is conducted by using an oxidizing agent commonly employed such as permanganic acid, periodic acid, peracetic acid, performic acid, trifluoroperacetic acid, perbenzoic acid, m-chloroperbenzoic acid, hydrogen peroxide, etc. The oxidizing agent can be used in large excess, and may be employed preferably in an amount of about 1 to about 2 moles per mole of the starting compound. The oxidation is carried out usually in a suitable solvent. Useful solvents include any of those which do not adversely affect the oxidation reaction such as chloroform, pyridine, tetrahydrofuran, dioxane, methylene chloride, carbon tetrachloride, acetic acid, formic acid, dimethylformamide, water, etc. The oxidation is performed at a temperature which is not particularly limited but generally ranges from room temperature to cooling temperature, preferably about 0° to about 30° C.

The compound thus obtained is subjected to de-esterification whereby the compound of the formula (II) can be produced. The de-esterification is effected under the same conditions as shown in the reaction scheme of the step (B). The process for preparing the compound of the formula (II) is described in detail in reference examples to be set forth later.

Reaction Equation-2



In the foregoing formulae, R_4 is as defined above, R_1' and R_5' are the same groups as those represented by R_1 and R_5 and at least one of them is trialkylsilyl group, and R_6 represents hydrogen or $COOR_2'$ wherein R_2' is as defined above.

The compound of the formula (I) wherein at least one of R_1 and R_2 is hydrogen atom, namely the compound of the formula (I-b), can be prepared by the process shown above in Reaction Equation-2. The steps in the process are set forth below in detail.

Step (C)

The compound of the formula (II) is reacted with a compound of the formula (III') in a solvent such as dichloromethane, dichloroethane, chloroform or like halogenated hydrocarbons. During this reaction, reaction for removing the trialkylsilyl group proceeds at the same time, whereby a compound of the formula (VI) is produced. Useful solvents are not particularly limited as far as they are halogenated hydrocarbons. The reaction conditions including the reaction temperature, the proportions of the reagents to be used and the reaction time are similar to those in the step (A).

Depending upon the kind of the penicillin carboxyl-protecting group represented by R_4, the compound of the formula (VI) thus obtained may be the product as contemplated, i.e., an ester of the penicillin derivative of

the formula (I). More preferably the ester of the formula (VI) is subjected to de-esterification as in the step (B) so that the compound is transformed to a penicillin derivative of the present invention during the formula (I-b) in which R_3 is hydrogen which is converted, when required, in the conventional manner into a pharmaceutically acceptable salt thereof or ester ther contemplated

Step (D)

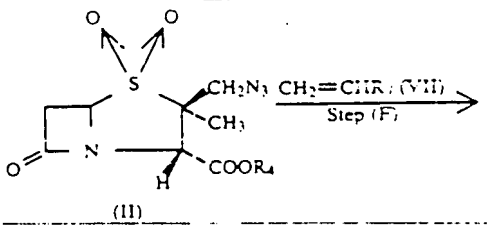
The compound of the formula (VI) is subjected to de-esterification after or without isolation from the reaction product obtained in the step (C), whereby a penicillin derivative of the formula (I-b) in which R_3 is hydrogen is produced. The de-esterification is carried out under the same conditions as those described above in respect of the step (B).

The compound of the formula (VI) can be prepared by the process in the step (C) and also by the process to be set forth below in step (E).

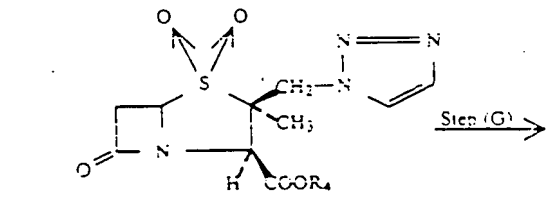
Step (E)

The compound of the formula (IV) obtained in the step (A) as shown in Reaction Equation-1 wherein at least one of R_1 and R_5 is trialkylsilyl, namely the compound of the formula (IV'), is subjected to reaction for removing the trialkylsilyl in the presence of potassium fluoride after or without isolation from the reaction product obtained in the step (A), whereby a compound of the formula (VI) is produced. The trialkylsilyl-removing reaction is conducted in a suitable solvent by using potassium fluoride in an amount of over about 1 mole, preferably about 1 mole, and a catalyst in an amount of about 1/50 to about 1/10 mole, both per mole of the compound of the formula (IV). Useful as the catalyst is a phase transfer catalyst such as quaternary ammonium salt, crown ether or the like. Examples of useful solvents are any suitable solvents which do not adversely affect the reaction and which include benzene, toluene, xylene or like aromatic hydrocarbons; acetonitrile, N,N-dimethylformamide, dimethylsulfoxide or like non-protonic polar solvents; etc. The reaction temperature and reaction time are appropriately determined. Generally the reaction is performed at a temperature in the range of room temperature to about 100° C., and completes in about 1 to about 10 hours.

Reaction Equation-3



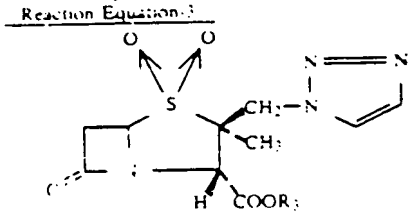
(II)



(VIII)

-continued

Reaction Equation-3



(I-c)

In the foregoing formulae, R_4 is as defined above, and R_7 represents acyloxy group.

Examples of the acyloxy groups represented by R_7 are lower acyloxy groups having 2 to 5 carbon atoms such as acetoxy, propionyloxy, butyryloxy, valeryloxy or like aliphatic acyloxy groups and benzoyloxy or like aromatic acyloxy groups, etc.

The compound of the formula (I) wherein R_1 and R_2 are hydrogen atoms, namely the compound of the formula (I-c), can be produced by the process as shown above in Reaction Equation-3.

The steps (F) and (G) in Reaction Equation-3 will be described below in detail.

Step (F)

The penicillanic acid derivative of the formula (II) is reacted with a vinyl derivative of the formula (VII) while reaction for removing the acyloxy group represented by R_7 in the formula (VII) is carried out, whereby a compound of the formula (VIII) is prepared. The reaction between the penicillanic acid derivative of the formula (II) and the vinyl derivative of the formula (VII) is conducted in the presence of or in the absence of a suitable solvent by using the vinyl derivative of the formula (VII) in an amount of at least about 1 mole, preferably from 1 to about 200 moles, per mole of the derivative of the formula (II), whereby there occurs simultaneously the acyloxy-removing reaction. The solvents which can be used are not particularly limited as far as they do not adversely affect the reaction. Specific examples thereof are benzene, toluene, xylene or like aromatic hydrocarbons, tetrahydrofuran, dioxane or like ethers, etc. The reaction is effected at a temperature ranging from about 50° C. to a boiling point of the solvent, or a temperature of less than 200° C. in a sealed reactor, and is completed in about 2 to about 72 hours. Depending on the kind of the penicillin carboxyl-protecting group represented by R_4 in the formula (VIII), the compound of the formula (VIII) thus obtained may be the product as contemplated, namely the ester of the penicillin derivative of the forumla (I). More preferably the compound of the formula (VIII) thus prepared is subjected to de-esterification as in the step (G) so that the compound is converted by the conventional method into a penicillin derivative of the formula (I-c) wherein R_3 is hydrogen which, in turn, is transformed by the conventional method into a pharmaceutically acceptable salt thereof or ester thereof as contemplated. The compound of the formula (VIII) can be made into a pharmaceutically acceptable salt thereof or ester thereof as contemplated by conducting an ester interchange or salt-forming reaction in the conventional manner.

Step (G)

The compound of the formula (VIII) is subjected to de-esterification after or without isolation from the

reaction product obtained in the step (F), whereby a penicillin derivative of the formula (I-c) in which R_3 is hydrogen is produced. The reaction conditions for de-esterification are the same as those described in the step (B).

After completion of the reaction in each step, the contemplated compound producible in each step can be isolated from the reaction product or, when required, can be purified by the conventional method such as recrystallization method, thin-layer chromatography, column chromatography or the like.

The penicillin derivative of the present invention is mixed with the β-lactam type antibiotic substance to form a preparation which is orally or parenterally administered. Alternatively, the present compound and a suitable antibiotic can be separately administered. Thus the derivatives of the formula (I) can be used for treating infectious disease of human beings and other animals.

The composition of the present invention may be made into tablets, pills, capsules, granules, powders, syrups, lozenges, solutions, suspensions, etc. for oral administration and aqueous, suspending or water-soluble preparations for intravenous, subcutaneous or intramuscular injections.

Carriers useful in formulating the preparations are commonly used pharmaceutically acceptable non-toxic carriers such as gelatin, lactose, starch, magnesium stearate, talc, vegetable oil, animal oil, polyalkylene glycol, etc. The carrier may be used with other additives such as diluents, binders, buffer agents, preservatives, glazes, disintegrators, coating agents, etc.

The daily dose of the preparation can be appropriately determined and is not particularly limited. Preferably the daily dose is such that the total amount of the present compound and β-lactam antibiotic is about 1 to about 200 mg/Kg body weight for oral administration and about 1 to about 100 mg/Kg body weight for parenteral administration.

The present invention will be described below in more detail with reference to examples given below.

REFERENCE EXAMPLE 1

Preparation of benzhydryl 2β-azidiomethyl-2α-methylpenam-3α-carboxylate

A solution of 5.00 g of sodium azide in 53 ml of water was added to a solution of benzhydryl 2β-chloromethyl-2α-methylpenam-3α-carboxylate (5.13 g) in dimethylformamide (155 ml). The mixture was stirred at room temperature for 4 hours. The resulting reaction mixture was poured into cooled water and the mixture was extracted with ethyl acetate. The ethyl acetate layer was washed with water, dried over magnesium sulfate and concentrated to provide 4.87 g of the contemplated product as oil in 93% yield.

Infrared absorption spectrum (nujol) νmax (cm^{-1}): 2120, 1812, 1765

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.30 (3H, s), 3.25 (2H, m), 3.42 (1H, d), 3.63 (1H, d), 4.75 (1H, s), 4.76 (1H, m), 7.00 (1H, s), 7.40 (10H, s)

REFERENCE EXAMPLE 2

Preparation of benzhydryl 2β-azidomethyl-2α-methylpenam-3α-carboxylate 1,1-dioxide

To a solution of benzhydryl 2β-azidomethyl-2α-methylpenam-3α-carboxylate (7.03 g) in a mixture of acetic acid (240 ml) and water (40 ml) was added potassium permanganate (6.02 g) over a period of more than 1 hour. The mixture was stirred at room temperature for 2.5 hours. The resulting reaction mixture was diluted with ice water. The precipitate was collected by filtration, and washed with water. The resulting product was dissolved in ethyl acetate and the solution was washed with an aqueous solution of sodium hydrogencarbonate and dried over magnesium sulfate. Concentration gave 5.48 g of the contemplated product in 72% yield.

Infrared absorption spectrum (nujol) νmax (cm^{-1}): 2120, 1812, 1765

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.18 (3H, s), 3.50 (2H, d), 3.72 (1H, d), 3.93 (1H, d), 4.60 (1H, m), 4.65 (1H, s), 7.00 (1H, s), 7.36 (10H, s)

REFERENCE EXAMPLE 3

Preparation of p-nitrobenzyl 2β-azidomethyl-2α-methylpenam-3α-carboxylate

The procedure of Reference Example 1 was repeated with the exception of using as the starting material p-nitrobenzyl 2β-chloromethyl-2α-methylpenam-3α-carboxylate, affording the above contemplated compound.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 2120, 1798, 1760

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.40 (3H, s), 3.12 (1H, dd), 3.50 (2H, s), 3.62 (1H, dd), 4.83 (1H, s), 5.29 (2H, s), 5.36 (1H, dd), 7.56 (2H, d), 8.26 (2H, d)

REFERENCE EXAMPLE 4

Preparation of p-nitrobenzyl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide

The procedure of Reference Example 2 was followed with the exception of using as the starting material p-nitrobenzyl 2β-azidomethyl-2α-methylpenam-3α-carboxylate, giving the above contemplated compound.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 2120, 1770

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.42 (3H, s), 3.45–3.60 (2H, m), 3.75 (1H, d), 3.96 (1H, d), 4.56–4.75 (1H, m), 4.64 (1H, s), 5.33 (2H, s), 7.56 (2H, d), 8.26 (2H, d)

EXAMPLE 1

Preparation of p-nitrobenzyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 1) and p-nitrobenzyl 2β-(5-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 2)

A 2.1 g quantity of p-nitrobenzyl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide and 0.63 g of ethyl propiolate in 62 ml of benzene were refluxed with stirring under nitrogen atmosphere for 37 hours. The solvent was removed by distillation and the residue was subjected to column chromatography on silica gel to produce as a first eluted product 0.7 g of p-nitrobenzyl

2β-(5-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in amorphous form (Compound 2) in 27% yield.

Infrared absorption spectrum (KBr) νmax (cm⁻¹): 1795, 1755, 1727

Nuclear magnetic resonance spectrum (CDCl₃) δ (ppm): 1.39 (3H, s), 1.39 (3H, t), 3.48–3.60 (2H, m), 4.42 (2H, q), 4.58–4.70 (1H, m), 5.11 (1H, s), 5.14 (1H, d), 5.25 (1H, d), 5.31 (1H, d), 5.56 (1H, d), 7.54 (2H, d), 8.09 (1H, s), 8.25 (2H, d).

There was obtained as a second eluted product 1.6 g of p-nitrobenzyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in amorphous form (Compound 1) in 62% yield.

Infrared absorption spectrum (KBr) νmax (cm⁻¹): 1800, 1760 (sh), 1733

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.34 (3H, s), 1.41 (3H, t), 3.50–3.65 (2H, m), 4.42 (2H, q), 4.60–4.75 (2H, m), 5.09 (2H, s), 5.36 (2H, s), 7.59 (2H, d), 8.28 (2H, d), 8.30 (1H, s)

EXAMPLE 2

Preparation of p-nitrobenzyl 2β-(4-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 3) and p-nitrobenzyl 2β-(5-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 4)

The contemplated product was synthesized in the same manner as in Example 1 and eluted by column chromatography on silica gel. There was obtained as a first eluted product p-nitrobenzyl 2β-(5-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in amorphous form (Compound 4) in 26% yield.

Infrared absorption spectrum (KBr) νmax (cm⁻¹): 1795, 1727

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.39 (3H, s), 3.45–3.60 (2H, m), 3.94 (3H, s), 4.58–4.70 (1H, m), 5.09 (1H, s), 5.10–5.64 (4H, m), 7.54 (2H, d), 8.10 (1H, s), 8.25 (2H, d).

There was obtained as a second eluted product p-nitrobenzyl 2β-(4-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in amorphous form (Compound 3) in 61% yield.

Infrared absorption spectrum (KBr) νmax (cm⁻¹): 1798, 1730

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.33 (3H, s), 3.48–3.68 (2H, m), 3.96 (3H, s), 4.56–4.76 (2H, m) 5.09 (2H, s), 5.36 (2H, s), 7.60 (2H, d), 8.28 (2H, d), 8.30 (1H, s)

EXAMPLE 3

Preparation of benzhydryl 2β-(4-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 5) and benzhydryl 2β-(5-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 6)

The contemplated product was synthesized in the same manner as in Example 1 and eluted by column chromatography on silica gel. First there was eluted benzhydryl 2β-(5-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 6) in 18% yield.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1800, 1727

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.20 (3H, s), 3.44–3.58 (2H, m), 3.91 (3H, s), 4.50–4.65 (1H, m), 5.24 (1H, d), 5.25 (1H, s), 5.45 (1H, d), 6.91 (1H, s), 7.20–7.40 (10H, m), 8.08 (1H, s).

Secondly there was eluted benzhydryl 2β-(4-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (compound 5) in 60% yield.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1803, 1727

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.05 (3H, s), 3.48–3.62 (2H, m), 3.95 (3H, s), 4.55–4.75 (2H, m), 5.11 (2H, bs), 7.02 (1H, s), 7.20–7.50 (10H, m), 8.25 (1H, s).

EXAMPLE 4

Preparation of sodium 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 7)

Hydrogenation was conducted at a low pressure and at room temperature by using 15 ml of ethyl acetate, 15 ml of water, 340 mg of p-nitrobenzyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide, 60 mg of 10% palladium charcoal and 110 mg of sodium hydrogencarbonate. After completion of absorption of hydrogen, the reaction mixture was filtered to separate the aqueous layer which was washed with benzene. The aqueous solution was concentrated at reduced pressure and the concentrate was subjected to column chromatography using an MCI gel, CHP-20 P (product of Mitsubishi Kasei Co., Ltd., Japan) to conduct gradient elution with a water-10% acetone water mixture. The eluate thus obtained was freeze-dried to afford 200 mg of the contemplated product (Compound 7) as white powder in 76% yield. The white powder decomposed at a temperature of more than 180° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1782, 1720

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.39 (3H, t), 1.46 (3H, s), 3.45 (1H, dd), 3.72 (1H, dd), 4.44 (2H, q), 4.50 (1H, s), 4.96–5.10 (1H, m), 5.18 (1H, d), 5.42 (1H, d), 8.72 (1H, s)

EXAMPLE 5

Preparation of 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylic acid-1,1-dioxide (Compound 8)

Hydrogenation was conducted at room temperature and at a pressure of 3 atm. by using 4.2 g of p-nitrobenzyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide, 1.4 g of sodium hydrogencarbonate, 800 mg of 10% palladium charcoal, 100 ml of ethyl acetate and 100 ml of water. After completion of absorption of hydrogen, the reaction mixture was filtered and the aqueous layer was separated and washed with benzene. The pH of the aqueous layer was adjusted to 1 to 2 with hydrochloric acid. The aqueous layer was extracted with ethyl acetate and the extract was dried over magnesium sulfate. The solvent was distilled off and 3.0 g of the contemplated compound was produced in amorphous form in 97% yield.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1798, 1726

Nuclear magnetic resonance spectrum (DMSO-d_6) δ (ppm): 1.31 (3H, t), 1.42 (3H, s), 3.31 (1H, dd), 3.73 (1H dd), 4.32 (2H, q), 4.75–5.38 (4H, m), 8.76 (1H, s)

EXAMPLE 6

Preparation of chloromethyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 9)

A 2.2 g quantity of sodium hydrogencarbonate and 0.2 g of tetrabutylammonium hydrogensulfate were added with stirring at a temperature of less than 10° C. to 2.4 g of 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylic acid-1,1-dioxide, 13.5 ml of dichloromethane and 13.5 ml of water. To the mixture was dropwise added at the same temperature 1.25 g of chloromethyl chlorosulfonate and the resulting mixture was stirred at room temperature for 30 minutes. The organic layer was separated, washed once with water and dried over magnesium sulfate. The solvent was removed by distillation and the residue was purified by column chromatography on silica gel, giving 2.2 g of the contemplated compound in amorphous form in 81% yield.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1798, 1723

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.42 (3H, t), 1.48 (3H, s), 3.52–3.65 (2H, m), 4.36 (2H, q), 4.60–4.78 (2H, m), 5.10 (2H, s), 5.73 (1H, d), 5.90 (1H, d), 8.31 (1H, s)

EXAMPLE 7

Preparation of iodomethyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 10)

A 1.73 g quantity of chloromethyl 2β-(4-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylic acid-1,1-dioxide and 1.3 g of sodium iodide were stirred in 3.4 ml of acetone at room temperature for 18 hours. To the reaction mixture was added 2.9 ml of water and the pH of the resulting mixture was adjusted to 7 to 8 with an aqueous solution of sodium hydrogencarbonate. After addition of 2.9 ml of water, the mixture was decolorized with an aqueous solution of 0.5M sodium thiosulfate, extracted with dichloromethane, washed with water and dried over magnesium sulfate. The solvent was removed by distillation and 1.9 g of the contemplated compound was prepared in amorphous form in 90% yeild.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1798, 1725

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.43 (3H, t), 1.49 (3H, s), 3.52–3.68 (2H, m), 4.43 (2H, q), 4.59–4.78 (2H, m), 5.09 (2H, s), 5.96 (1H, d), 6.07 (1H, d), 8.32 (1H, s)

EXAMPLE 8

Preparation of sodium 2β-(5-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 11)

A 220 mg of the contemplated compound was prepared in the form of white powder in the same manner

as in Example 4 from 0.34 g of p-nitrobenzyl 2β-(5-ethoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in 83% yield.

The white powder thus obtained decomposed at a temperature of over 180° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1788, 1736

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.39 (3H, t), 1.43 (3H, s), 3.40 (1H, dd), 3.71 (1H, dd), 4.46 (2H, q), 4.57 (1H, s), 4.96–5.05 (1H, m), 5.40 (1H, d), 5.82 (1H, d), 8.34 (1H, s)

EXAMPLE 9

Preparation of sodium 2β-(4-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 12)

A 0.18 g quantity of the contemplated product was prepared as white powder in the same manner as in Example 4 from 0.3 g of p-nitrobenzyl 2β-(4-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in 78% yield.

The white powder thus obtained decomposed at a temperature of over 184° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1782, 1730

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.46 (3H, s), 3.45 (1H, dd), 3.73 (1H, dd), 3.97 (3H, s), 4.50 (1H, s), 4.81 (2H, s), 4.98–5.10 (1H, m), 5.18 (1H, d), 5.42 (1H, d), 8.72 (1H, s)

EXAMPLE 10

Preparation of sodium 2β-(5-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 13)

A 0.19 g quantity of the contemplated compound was prepared as white powder in the same manner as in Example 4 from 0.3 g of p-nitrobenzyl 2β-(5-methoxycarbonyl-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide in 82% yield.

The white powder thus obtained decomposed at a temperature of over 180° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1778, 1730

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.41 (3H, s), 3.41 (1H, dd), 3.71 (1H, dd), 3.98 (3H, s), 4.56 (1H, s), 4.95–5.08 (1H, m), 5.40 (1H, d), 5.83 (1H, d), 8.34 (1H, s)

EXAMPLE 11

Preparation of p-nitrobenzyl 2α-methyl-2β-[4-(p-nitrobenzyloxycarbonyl)-1,2,3-triazol-1-yl]methylpenam-3α-carboxylate-1,1-dioxide (Compound 14) and p-nitrobenzyl 2α-methyl-2β-[5-(p-nitrobenzyloxycarbonyl)-1,2,3-triazol-1-yl]methylpenam-3α-carboxylate-1,1-dioxide (Compound 15)

A 4 g quantity of p-nitrobenzyl 2β-adidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide and 8.2 g of p-nitrobenzyl acetylene carboxylate in 100 ml of benzene were refluxed under nitrogen atmosphere for 12 hours. The solvent was distilled off at reduced pressure. The residue was subjected to column chromatography on silica gel to provide 3.6 g of p-nitrobenzyl 2α-methyl-2β-[4-(p-nitrobenzyloxycarbonyl)-1,2,3-triazol-1-yl]methylpenam-3α-carboxylate-1,1-dioxide (Com-

pound 14) and 0.9 g of p-nitrobenzyl 2α-methyl-2β-[5-(p-nitrobenzyloxycarbonyl)-1,2,3-triazol-1-yl]methylpenam-3α-carboxylate-1,1-dioxide (Compound 15) both in amorphous form.

Compound 14

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1800, 1740

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.34 (3H, s), 3.3–3.6 (2H, m), 4.57 (1H, s), 4.60–4.76 (1H, m) 5.12 (2H, s), 5.37 (2H, s), 5.48 (2H, s), 7.5–7.7 (4H, m), 8.1–8.3 (4H, m), 8.37 (1H, s).

Compound 15

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1800, 1740

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.41 (3H, s), 3.3–3.7 (2H, m), 4.6–4.7 (1H, m), 5.07 (1H, s), 5.1–5.6 (4H, m), 5.46 (2H, s), 7.4–7.7 (4H, m), 8.15 (1H, s), 8.1–8.4 (4H, m)

EXAMPLE 12

Preparation of dipotassium 2β-(4-carboxy-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 16)

Hydrogenation was conducted in 100 ml of ethyl acetate and 100 ml of water at room temperature for 1 hour by using 3.6 g of p-nitrobenzyl 2α-methyl-2β-[4-(p-nitrobenzyloxycarbonyl)-1,2,3-triazol-1-yl)]methylpenam-3α-carboxylate-1,1-dioxide, 2.0 g sodium hydrogencarbonate and 0.68 g of 10% palladium charcoal, catalyst. Thereafter the aqueous layer was separated and was washed once with ethyl acetate, and the pH thereof was adjusted to 1.5 to 1.7 with 6N hydrochloric acid. The aqueous solution was saturated with sodium chloride and extracted a few times with ethyl acetate. The ethyl acetate solutions thus formed were collected and dried over magnesium sulfate. The solvent was distilled off at reduced pressure to provide as the residue a foamed product of 2β-(4-carboxy-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylic acid-1,1-dioxide.

A 2 g quantity of the 2β-(4-carboxy-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylic acid-1,1-dioxide was dissolved in 20 ml of butanol. To the solution was added a solution of potassium 2-ethyl hexanoate in butanol, and the mixture was stirred awhile at room temperature. The precipitate was filtered to give 2.0 g of white solids having a melting point of over 178° C. (decomposition).

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1780, 1610

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.47 (3H, s), 3.49 (1H, dd), 3.77 (1H, dd), 4.53 (1H, s), 5.0–5.1 (1H, m), 5.16 (1H, d), 5.41 (1H, d), 8.47 (1H, s)

EXAMPLE 13

Preparation of dipotassium 2β-(5-carboxy-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 17)

White solid of the contemplated compound with a melting point of over 175° C. (decomposition) was prepared in the same manner as in Example 12 by using p-nitrobenzyl 2α-methyl-2β-[5-(p-nitrobenzyloxycarbonyl)-1,2,3-triazol-1-yl]methylpenam-3α-carboxylate-1,1-dioxide.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1780, 1610

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.40 (3H, s), 3.43 (1H, dd), 3.71 (1H, dd), 4.58 (1H, s), 4.9–5.1 (1H, m), 5.36 (1H, d), 5.93 (1H, d), 8.04 (1H, s)

EXAMPLE 14

Preparation of benzhydryl 2β-(4-carboxy-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 18)

A 0.5 g quantity of benzhydryl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide and 0.083 g of acetylenecarboxylic acid were stirred in 2 ml of dichloromethane at room temperature under nitrogen atmosphere for 24 hours. The solvent was removed by distillation at reduced pressure and to the residue oil was added benzene. The insolubles were filtered off and to the residue was added hexane to deposit crystals which were collected by filtration. Thus there was produced 0.23 g of white crystals which melt at 120° to 121° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1805, 1745

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.07 (3H, s), 3.2–3.8 (2H, m), 4.5–4.7 (1H, m), 4.69 (1H, s), 5.12 (2H, bs), 7.02 (1H, s), 7.1–7.6 (10H, m), 8.33 (1H, s)

EXAMPLE 15

Preparation of disodium 2β-(4-carboxly-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide (Compound 19)

Hydrogenation was conducted in 10 ml of ethyl acetate and 10 ml of water at room temperature for 30 minutes by using 49 mg of benzhydryl 2β-(4-carboxly-1,2,3-triazol-1-yl)methyl-2α-methylpenam-3α-carboxylate-1,1-dioxide, 15 ml of 10% palladium charcoal and 24 mg of sodium hydrogencarbonate. The aqueous layer was separated from the reaction mixture and washed with ethyl acetate, and was purified with an MCI gel, CHP-20P (product of Mitsubishi Kasei Co., Ltd., Japan). After freeze-drying, there was obtained a white amorphous product having a melting point of 220° to 250° C. (decomposition).

The values of the infrared absorption spectrum and nuclear magnetic resonance spectrum of the compound thus obtained were similar to those of Compound 16 prepared in Example 12.

EXAMPLE 16

Preparation of benzhydryl 2α-methyl-2β-(4-trimethylsilyl-1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 20)

A 150 mg quantity of benzhydryl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide was reacted in a sealed reactor with 300 mg of trimethylsilylacetylene at 90° to 95° C. for 20 hours. The reaction mixture was concentrated at reduced pressure, giving 170 mg of white crystals which melt at 172° to 175° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1805, 1755

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 0.32 (9H, s), 1.05 (3H, s), 3.3–3.7 (2H, m), 4.5–4.7 (1H, m), 4.65 (1H, s), 5.08 (2H, AB-q), 7.00 (1H, s), 7.3–7.5 (10H, m), 7.67 (1H, s)

EXAMPLE 17

Preparation of benzhydryl 2α-methyl-2β-(1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 21)

A 133 mg quantity of benzhydryl 2α-methyl-2β-(4-trimethylsilyl-1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide, 3.26 g of 18-crown-6(1,4,7,10,13,16-hexaoxacyclooctadecane) and 15.8 mg of potassium fluoride were stirred in 0.7 ml of N,N-dimethylformamide at 50° to 60° C. for 5.5 hours. The reaction mixture was poured into excess iced water and the mixture was extracted a few times with ethyl acetate. The ethyl acetate extracts were collected and dried over magnesium sulfate. The solvent was distilled off at reduced pressure and the residue was purified by column chromatography on silica gel, whereby a white product was given which has a melting point of 206° to 208° C. (decomposition).

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1800, 1760

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.05 (3H, s), 3.3–3.7 (2H, m), 4.5–4.7 (1H, m), 4.65 (1H, s), 5.10 (2H, AB-q), 7.00 (1H, s), 7.3–7.5 (10H, m), 7.73 (1H, s)

EXAMPLE 18

Preparation of benzhydryl 2α-methyl-2β-(1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 21)

A 500 mg quantity of benzhydryl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide, 335 mg of trimethylsilylacetylene and 2 ml of methylene chloride were reacted in a sealed reactor at 95° C. for 20 hours. The reaction mixture was concentrated at reduced pressure and the residue was purified by column chromatography on silica gel to provide white solids having a melting point of 203° to 204° C. (decomposition).

Fast atomic bombardment mass spectrum method: m/e = 467(M^+)

The values of the infrared absorption spectrum and nuclear magnetic resonance spectrum of te compound thus obtained were identical with those of Compound 21 obtained in Example 17.

EXAMPLE 19

Preparation of benzhydryl 2α-methyl-2β-(1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 21)

A 200 mg quantity of benzhydryl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide was reacted with 10 ml of vinyl acetate in a sealed reactor at 100° to 110° C. for 30 hours. The reaction mixture was concentrated at reduced pressure. The residue was crystallized with cooled chloroform.

The white crystals thus obtained were found to have a melting point (decomposition) and the values of the nuclear magnetic resonance spectrum which were all identical with the values of Compound 21 obtained in Example 17.

EXAMPLE 20

Preparation of sodium 2α-methyl-2β-(1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 22)

Hydrogenation was conducted in 10 ml of ethyl acetate and 10 ml of water at room temperature for 30 minutes by using 45 mg of benzhydryl 2α-methyl-2β-(1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide, 15 mg of 10% palladium charcoal and 16 mg of sodium hydrogencarbonate. The aqueous layer was separated from the reaction mixture and washed once with ethyl acetate. The aqueous solution was then purified with an MCI gel, CHP-20P (product of Mitsubishi Kasei Co., Ltd., Japan). After freeze-drying, there was obtained an amorphous product with a melting point of over 170° C. (decomposition).

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1780, 1630

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 1.41 (3H, s), 3.45 (1H, dd), 3.72 (1H, dd), 4.48 (1H, s), 4.96–5.10 (1H, m), 5.25 (2H, AB-q), 7.85 (1H, d), 8.13 (1H, d)

EXAMPLE 21

Preparation of p-nitrobenzyl 2α-methyl-2β-(1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 23)

A 1.02 g quantity of p-nitrobenzyl 2β-azidomethyl-2α-methylpenam-3α-carboxylate-1,1-dioxide was reacted with 50 ml of vinyl acetate in a sealed reactor at 100° to 110° C. for 30 hours. The reaction mixture was concentrated at reduced pressure and the residue was purified by column chromatography on silica gel, giving 0.73 g of the contemplated compound in amorphous form in 67% yield which melts at 182° to 184° C.

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1800, 1760

Nuclear magnetic resonance spectrum ($CDCl_3$) δ (ppm): 1.26 (3H, s), 3.5–3.6 (2, Hm), 4.66 (1H, s), 4.6–4.7 (1H, m) 5.07 (2H, s), 5.36 (2H, s), 7.61 (2H, d), 7.74 (1H, d), 7.80 (1H, d), 8.28 (2H, d)

EXAMPLE 22

Preparation of sodium 2α-methyl-2β-(4-trimethylsilyl-1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide (Compound 24)

Hydrogenation was performed in 15 ml of ethyl acetate and 15 ml of water at room temperature for 30 minutes by using 200 mg of benzhydryl 2α-methyl-2β-(4-trimethylsilyl-1,2,3-triazol-1-yl)methylpenam-3α-carboxylate-1,1-dioxide, 50 mg of 10% palladium charcoal and 98 mg of sodium hydrogencarbonate. The aqueous layer was removed from the reaction mixture and washed once with ethyl acetate. The aqueous solution was purified with an MCI gel, CHP-20P (product

of Mitsubishi Kasei Co., Ltd., Japan) After freeze-drying, there was obtained an amorphous product having a melting point of over 170° C. (decomposition).

Infrared absorption spectrum (KBr) νmax (cm^{-1}): 1780, 1630

Nuclear magnetic resonance spectrum (D_2O) δ (ppm): 0.32 (9H, s), 1.38 (3H, s), 3.1-3.7 (2H, m), 4.46 (1H, s), 4.9-5.0 (1H, m), 5.23 (2H, AB-q), 8.16 (1H, s)

The compounds obtained in some of the examples were checked for β-lactamase inhibitory activity and antibacterial activity.

(1) Test for β-lactamase inhibitory activity

The inhibitory activity against penicillinase (β-lactamase) from Bacillus SP was measured by microiodometry Tanpakushitsu Kakusan Koso (Protein Nucleic Acid Enzyme), vol. 23, No. 5, pp 391–400 (1978) using a penicillin G as a substrate. Table 1 given below shows the results.

TABLE 1

Compound	50% Inhibitory Concentration
Compound 7	2.4×10^{-8} M
Compound 11	3.4×10^{-7} M
Compound 12	4.9×10^{-8} M
Compound 13	3.0×10^{-7} M
Compound 5	6.0×10^{-7} M
Compound 17	1.7×10^{-6} M
Compound 22	6.9×10^{-7} M
Compound 24	5.1×10^{-7} M

(2) Test for antibacterial activity

(1) Effects by ampicillin as combined with the present compound

The compounds of the present invention and ampicillin, each singly used, were checked for minimal inhibitory concentration (MIC) against the bacteria listed in Table 2 given below by micro-broth dilution method ("American Journal Clinical Pathology" published in 1980, vol. 73, No. 3, pp 374 to 379). The MIC of ampicillin as combined with the present compound (10 μg/ml) was measured against the same bacteria. In the method, the bacteria cultivated in Mueller Hinton Broth (product of DIFCO) and diluted to 10^7 CFU/ml were inoculated into the same medium containing ampicillin and the present compound in a specific concentration, and incubated at 37° C. for 20 hours. Thereafter the growth of the microorganisms was observed to determine the minimal inhibitory concentration (MIC) for rendering the inoculated medium free from turbidity. The present compounds, singly used, turned out to be all more than 25 μg/ml in MIC. The bacteria as used in the test were those capable of producing β-lactamase, among which the bacteria marked * in the table are those collected from the living body of human hosts and the others are a stock culture.

In Table 2, the present compounds are shown by the compound number.

TABLE 2

Test Bacteria	MIC (μg/ml)								
	Ampicillin (singly used)	Present Compound (combined with ampicillin)							
		7	11	12	13	16	17	22	24
S. aureus S-54	25	0.1	0.2	0.2	0.2	0.2	0.78	0.2	0.78
S. aureus ATCC 90124	25	0.1	0.2	0.2	0.2	0.2	0.78	0.1	0.39
E. coli TH-13*	400	6.25	25	3.13	6.25	6.25	0.05	3.13	100
E. coli TH-397*	400	6.25	12.5	3.13	6.25	3.13	6.25	6.25	50

TABLE 2-continued

Test Bacteria	MIC (μg/ml) Ampicillin (singly used)	Present Compound (combined with ampicillin) 7	11	12	13	16	17	22	24
P. mirabilis 121	400	1.56	0.78	0.78	0.78	0.78	0.39	0.78	25
P. vulgaris HD OX-19	100	0.78	0.78	0.39	0.39	1.56	1.56	0.78	1.56
S. marcescens TH-05*	400	12.5	25	12.5	25	6.25	1.56	3.13	100

(2) Effects by antibiotics as combined with the present compound

The compounds of the present invention, ampicillin, mecillinam, piperacillin and cephalexin, each singly used, were also tested for minimal inhibitory concentration against 30 strains of coliform bacilli collected from the living body of humans. The MIC of each antibiotic as combined with the present compound (10 μg/ml) was likewise measured. Table 3 to 6 indicate the results in which MIC_{50} and MIC_{70} indicate the minimal inhibitory concentration for inhibiting the growth of 50% and 70% respectively of the strains. The MICs of the present compounds singly used were all more than 25 μg/ml.

TABLE 3

30 Strains of *coriform bacilli*	Ampicillin singly used	Present compound as combined with ampicillin Comp. 7	Comp. 11	Comp. 16	Comp. 17	Comp. 22
MIC_{50} (μg/ml)	400	6.25	50	6.25	25	3.13
MIC_{70} (μg/ml)	400	50	100	6.25	100	6.25

TABLE 4

30 Strains of *coriform bacilli*	Mecillinam singly used	Present compound as combined with mecillinam Comp. 7	Comp. 11	Comp. 16	Comp. 17	Comp. 22
MIC_{50} (μg/ml)	3.13	0.2	0.2	0.1	0.05	0.1
MIC_{70} (μg/ml)	12.5	0.39	0.39	0.1	0.39	0.2

TABLE 5

30 Strains of *coriform bacilli*	Piperacillin singly used	Present compound as combined with piperacillin Comp. 7	Comp. 11	Comp. 16	Comp. 17	Comp. 22
MIC_{50} (μg/ml)	50	1.56	6.25	1.56	6.25	1.56
MIC_{70} (μg/ml)	200	6.25	25	3.13	50	1.56

TABLE 6

30 Strains of *coriform bacilli*	Cephalexin singly used	Present compound as combined with cephalexin Comp. 7	Comp. 11	Comp. 16	Comp. 17	Comp. 22
MIC_{50} (μg/ml)	25	12.5	12.5	6.25	3.13	12.5
MIC_{70} (μg/ml)	100	100	100	25	12.5	50

Given below are examples of preparation of the present antibacterial compositions.

Preparation Example 1

Ampicillin	200 mg
Compound 22	200 mg
Lactose	100 mg
Crystalline cellulose	57 mg
Magnesium stearate	3 mg
Total	560 mg
	(amount per capsule)

The above ingredients are formulated in the proportions listed above into a capsule.

Preparation Example 2	
Amoxycillin	100 mg
Compound 16	70 mg
Lactose	330 mg
Corn starch	490 mg
Hydroxypropyl methyl cellulose	10 mg
Total	1000 mg (amount per dose)

The above ingredients are formulated in the proportions listed above into granules.

Preparation Example 3	
Pivmecillinam	70 mg
Compound 17	70 mg
Lactose	33 mg
Crystalline cellulose	15 mg
Magnesium stearate	3 mg
Talc	4 mg
Corn starch	15 mg
Hydroxypropyl methyl cellulose	10 mg
Total	220 mg (amount per tablet)

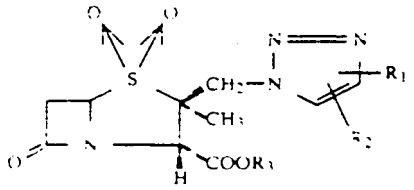
The above ingredients are formulated in the proportions listed above into a tablet.

Preparation Example 4	
Compound 22	120 mg
Hydroxypropyl cellulose	3 mg
Corn starch	25 mg
Magnesium stearate	2 mg
Total	150 mg
	(amount per tablet)

The above ingredients are formulated in the proportions listed above into a tablet.

We claim:

1. A penicillin derivative represented by the following formula



wherein R_1 is hydrogen or trialkylsily; R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt; and R_3 has the same meaning as above R_2'.

2. The penicillin derivative as defined in claim 1 wherein R_3 is C_{2-7} alkoxymethyl.

3. The penicillin derivative as defined in claim 1 wherein R_3 is C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl or C_{8-13} benzoyloxyalkyl.

4. The penicillin derivative as defined in claim 1 wherein R_3 is C_{3-8} alkoxycarbonylmethyl or C_{4-9} alkoxycarbonylethyl.

5. The penicillin derivative as defined in claim 1 wherein R_3 is phthalidyl.

6. The penicillin derivative as defined in claim 1 wherein R_3 is crotonolacton-4-yl and γ-butyrolacton-4-yl.

7. The penicillin derivative as defined in claim 1 wherein R_3 is (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl.

8. The penicillin derivative as defined in claim 1 wherein R_3 is a group for forming a pharmaceutically acceptable salt.

9. The penicillin derivative as defined in claim 1 wherein R_3 is C_{1-6} alkyl or halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylchlorosilyl and trichlorosilyl.

10. The penicillin derivative as defined in claim 8 wherein the group for forming a pharmaceutically acceptable salt represented by R_3 is alkali metal atom, alkaline earth metal atom or ammonium, or the group $COOR_3$ represents a carboxylic acid salt formed from

the carboxyl group and a member selected from the group consisting of cyclohexylamine, trimethylamine, diethanolamine, arginine and lysine.

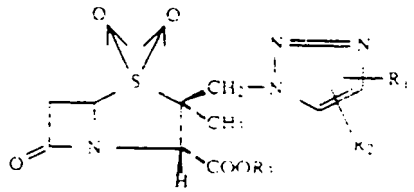
11. The penicillin derivative as defined in claim 1 wherein R_1 and R_2 are hydrogen.

12. The penicillin derivative as defined in claim 1 wherein R_1 is hydrogen and R_2 is $--COOR_2'$

13. The penicillin derivative as defined in claim 12 wherein R_2' is C_{1-18} alkyl.

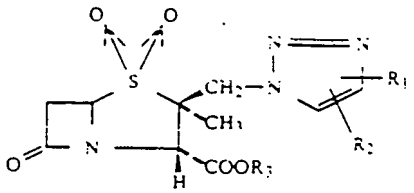
14. The penicillin derivative as defined in claim 1 wherein R_2 is trialkylsilyl.

15. A pharmaceutical composition useful for treating bacterial infections in mammals, said composition comprising (A) a β-lactam antibiotic and (B) a compound of the formula



wherein R_1 is hydrogen or trialkylsilyl; R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt; and R_3 has the same meaning as above R_2', the weight ratio of (A)/(B) being 0.1 to 10, said β-lactam antibiotics being selected from the group consisting of ampicillin, amoxicillin, hetacillin, ciclacillin, mecillinam, carbenicillin, sulbenicillin, ticarcillin, piperacillin, apalcillin, methicillin, mezlocillin, bacampicillin, carindacillin, talampicillin, carfecillin and pivmecillinam, cephaloridine, cephalothin, cephapirin, cephacetrile, cefazolin, cephalexin, cefradine, cefotiam, cefamandole, cefuroxime, cefoxitin, cefmetazole, cefsulodin, cefoperazone, cefotaxime, ceftizoxime, cefmenoxime, latamoxef, cefaclor, cefroxadine, cefatrizine, cefadroxil and cephaloglycin; and pharmaceutically acceptable salts thereof.

16. A method of treating a bacterial infection in a mammal subject, said method comprising administering to said subject (A) a β-lactam antibiotic and (B) a compound of the formula



wherein R_1 is hydrogen or trialkylsilyl; R_2 is hydrogen, trialkylsilyl or $COOR_2'$ wherein R_2' is hydrogen, C_{1-18} alkyl, C_{2-7} alkoxymethyl, C_{3-8} alkylcarbonyloxymethyl,

C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)carbonyloxymethyl, C_{9-13} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, halogenated C_{1-6} alkyl substituted with 1 to 3 halogen atoms, C_{1-6} alkoxy- or nitro-substituted or unsubstituted benzyl, benzhydryl, tetrahydropyranyl, dimethylaminoethyl, dimethylchlorosilyl, trichlorosilyl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt, and R_3 has the same meaning as above R_2', the weight ratio of (A)/(B) administered being 0.1 to 10, said β-lactam antibiotics being selected from the group consisting of ampicillin, amoxicillin, hetacillin, ciclacillin, mecillinam, carbenicillin, sulbenicillin, ticarcillin, piperacillin, apalcillin, methicillin, mezlocillin, bacampicillin, carindacillin, talampicillin, carfecilin and pivmecillinam, cephaloridine, cephalothin, cephapirin, cephacetrile, cefazolin, cephalexin, cefradine, cefotiam, cefamandole, cefuroxime, cefoxitin, cefmetazole, cefsulodin, cefoperazone, cefotaxime, ceftizoxime, cefmenoxime, fata-

moxef, cefaclor, cefroxadine, cefatrizine, cefadroxil and cephaloglycin; and pharmaceutically acceptable salts thereof.

17. The penicillin derivative as defined in claim 11 wherein R_3 is C_{3-8} alkylcarbonyloxymethyl, hydrogen, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)-carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt.

18. The penicillin derivative as defined in claim 12 wherein R_3 is C_{3-8} alkylcarbonyloxymethyl, hydrogen, C_{4-9} alkylcarbonyloxyethyl, (C_{5-7} cycloalkyl)-carbonyloxymethyl, C_{9-14} benzylcarbonyloxyalkyl, C_{3-8} alkoxycarbonylmethyl, C_{4-9} alkoxycarbonylethyl, phthalidyl, crotonolacton-4-yl, γ-butyrolacton-4-yl, (5-substituted C_{1-6} alkyl or phenyl or unsubstituted-2-oxo-1,3-dioxoden-4-yl)methyl, C_{8-13} benzoyloxyalkyl or group for forming a pharmaceutically acceptable salt

ATTACHMENT 2

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of: RONALD G. MICETICH et al
Attorney Docket: SAE-22
Serial Number: 519,491
Group Art Unit: 122
Filed: August 1, 1983
Examiner: N. Rizzo
For: PENICILLIN DERIVATIVES AND PROCESS FOR PREPARATION OF THE SAME

Date:

TERMINAL DISCLAIMER

The Hon. Commissioner of Patents
and Trademarks
Washington, D.C. 20231

Sir:

The Assignee of the above-identified United States patent application hereby disclaims any terminal portion of the patent to be issued from the above-identified application, which terminal portion exceeds the expiration date of any patent issued from commonly-assigned United States patent application serial number 501,560, filed June 6, 1983 in the names of Micetich, et al.

The Assignee herein disclaims any remaining term of any patent issued from the above-identified application if that patent should cease to be commonly-owned by the owners of any patent to issue from the said United States patent application serial number 501,560.

The Assignee herein is the Assignee of record as evidenced by the Assignment from the applicants recorded in the United States Patent and Trademark Office on August 1, 1983 at Reel 4161 Frames 964 and 965.

Taiho Pharmaceutical Company Ltd.

Assignee
By _____
Name Yukio Kobayashi
Title President
Date April 11, 1985

ATTACHMENT 3

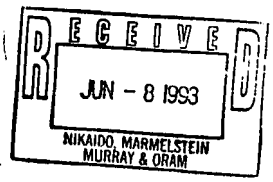
P102-1677

UNITED STATES DEPARTMENT OF COMMERCE
Patent and Trademark Office
Address: COMMISSIONER OF PATENTS AND TRADEMARKS
Washington, D. C. 20231

PAYOR NUMBER:
002841

75N3/0404
NIKAIDO, MARMELSTEIN, MURRAY & ORAM
655 FIFTEENTH STREET, N.W.
SUITE 330-S SUBLET LOBBY
WASHINGTON, DC 20005-5701

MAIL DATE:
05/28/93



MAINTENANCE FEE STATEMENT

The data shown below is from the records of the Patent and Trademark Office. If the maintenance fees and any necessary surcharges have been timely paid for the patents listed below, the notation "PAID" will appear in column 10, "status" below.

If a maintenance fee payment is defective, the reason is indicated by code in column 10, "status" below. An explanation of the codes appears on the reverse of the Maintenance Fee Statement. TIMELY CORRECTION IS REQUIRED IN ORDER TO AVOID EXPIRATION OF THE PATENT. NOTE 37 CFR 1.377. THE PAYMENT(S) WILL BE ENTERED UPON RECEIPT OF ACCEPTABLE CORRECTION. IF PAYMENT OR CORRECTION IS SUBMITTED DURING THE GRACE PERIOD, A SURCHARGE IS ALSO REQUIRED. NOTE 37 CFR 1.20(k) and (l).

If the statement of small entity status is defective the reason is indicated below in column 10 for the related patent number. THE STATEMENT OF SMALL ENTITY STATUS WILL BE ENTERED UPON RECEIPT OF ACCEPTABLE CORRECTION.

ITM NBR	PATENT NUMBER	PFC CDE	FEE AMOUNT	SUR CHARGE	SERIAL NUMBER	PATENT DATE	FILE DATE	PAY SML ENT STA
1	4,562,049	131	1370	-	06/519,491	12/31/85	05 01 82	08 190 PAI

If the "status" column for a patent number listed above does not indicate "PAID" a code or an asterisk (*) will appear in the "status" column. Where an asterisk (*) appears, the codes are set out below by the related item number. An explanation of the codes indicated in the "status" column and as set out below by the related item number appears on the reverse of the maintenance fee statement.

ITM STA

UNITED STATES DEPARTMENT OF COMMERCE
Patent and Trademark Office
COMMISSIONER OF PATENTS AND TRADEMARKS

[illegible] NUMBER
002841

ARMSTRONG, NIDAIDO, MARMELSTEIN,
KUBOVCIK & MURRAY
1725 K STREET, N.W.
SUITE 1000
WASHINGTON, DC 20006

DATE MAILED
04/22/89

MAINTENANCE FEE STATEMENT

The data shown below is from the records of the Patent and Trademark Office. If the maintenance fees and any necessary surcharges have been received for the patents listed below, the notation "PAID" will appear in column 10, "status" below.

If a maintenance fee payment is defective, the reason is indicated by code in column 10, "status" below. An explanation of the codes appears on the reverse of this Maintenance Fee Statement. TIMELY CORRECTION IS REQUIRED IN ORDER TO AVOID EXPIRATION OF THE PATENT. NOTE 37 CFR 1.377. THE PAYMENT(S) WILL BE ENTERED UPON RECEIPT OF ACCEPTABLE CORRECTION. IF PAYMENT OR CORRECTION IS SUBMITTED [illegible] THE GRACE PERIOD, A SURCHARGE IS ALSO [illegible] CFR [illegible]

[illegible] status [illegible] indicated below in column 10 for the [illegible] ENTERED UPON RECEIPT OF ACCEPTABLE CORRECTION.

ITM NBR	PATENT NUMBER	FEE AMOUNT	FEE CODE	SUR CHARGE	SERIAL NUMBER	PATENT DATE	FILE DATE	PAY YR	EXT	STA
1	4,558,[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	01/25/84	04	YES	PAI
2	4,560,[illegible]	[illegible]	150	[illegible]	[illegible]	12/24/[illegible]	11/19/84	04	NO	PAI
3	4,560,[illegible]	[illegible]	150	[illegible]	[illegible]	12/24/[illegible]	06/11/84	04	NO	PAI
4	4,562,[illegible]	[illegible]	150	[illegible]	[illegible]	[illegible]	08/01/83	04	NO	PAI

If the "status" column for a patent number listed above does not indicate "PAID" a code or an asterisk (*) will appear in the "status" column. Where an asterisk (*) appears, the codes are set out below by the related item number. An explanation of the codes indicated in the "status" column and as set out below by the related item number appears on the reverse of the maintenance fee statement.

ITM NBR	ATTY DKT NUMBER
1	P102-[illegible]-8404
2	P102-[illegible]-A868
3	P102-[illegible]-A868
4	SAE-22

DIRECT THE RESPONSE TOGETHER WITH [illegible]

ATTACHMENT 4

ZOSYN NDA 50-684 PATENT TIME CHRONOLOGY

DATE	EVENT
	IND ACTIVITIES
6/10/88	IND 31,705 filed with the Anti-Infective Division
7/29/88	FDA Meeting re initial clinical program.
8/24/88	FDA Meeting re preclinical and clinical issues.
10/27/88	Amendment for Protocol 68-5 Hospital Acquired Pneumonia
11/22/88	Amendment for Protocol 68-9 Skin and Skin Structure Infections
12/7/88	Amendment for Protocol 68-17 Intra-Abdominal Infections
2/23/89	FDA Meeting re Phase 1/ Pharmacokinetic Program
5/23/89	FDA Meeting re study 68-36, treatment of hospital acquired lower respiratory tract infections. Interim safety data presented.
5/30/89	IND Amendment containing mutagenicity and Segment 1 and 3 data filed.
6/6/89	FDA Meeting re toxicology and clinical issues; FDA requests added mutagenicity and repro testing. Gynecological Infection protocol discussed
7/27/89	FDA Meeting re format and content of Human PK and Biopharm NDA Section.
9/27/89	Amendement for Protocol 68-28 Gynecological Infections.
11/17/89	End of Phase II FDA Meeting focusing on safety aspects of clinical program. Neutropenia study discussed.
12/7/89	FDA Telecon with Ms. Tricia DeSantis, Project Manager, re 3 day reports.
1/30/90	Amendment for Protocol 68-36 Community Acquired Pneumonia.
2/20/90	Tox Amendment - 6 month rat IP and dog IV; 2 week dog SC pilot study as requested at 6/6/89 FDA meeting.
3/1/90	FDA Meeting re inital plans for Computer Assisted NDA (CANDA).
3/2/90	FDA Meeting re PK in renally impaired patients and M1 metabolite of tazobactam. Added data requested. NDA requirements for foreign studies discussed.
4/4/90	Additional mutagenicity studies submitted.

5/18/90	FDA Meeting re animal and human data supporting use in renally impaired patients to include 3 week dog tox study of M1 metabolite.
6/11/90	Reproductive toxicology studies in the rat submitted.
11/5/90	Submission of data supporting initiation of renal impairment study, including tox data on M1 metabolite.
1/9/91	FDA Pre-NDA Meeting; FDA willing to accept added "RS" pathogens from European trials. Format of study reports and "master table" (listings) discussed.
1/31/91	FDA removes hold on enrollment of renally impaired patients in multiple dose PK study.
2/13/91	FDA Pre-NDA Meeting to discuss filing strategy. Microbiology, data conventions and evaluability were also discussed.
4/19/91	FDA Meeting re Biopharm CANDA and content/format of Human PK and Biopharm NDA Section.
5/1/91	FDA Meeting re design of CANDA for use by medical reviewer.
5/17/91	FDA telecon re design of CANDA for use by statistical reviewer.

NDA Activities

8/30/91	NDA 50-684 filed with Anti-Infective Division for Intra-Abdominal and Skin and Skin Structure
2/28/92	NDA Amendment for Gynecological Infections
4/30/92	NDA Amendment for Community Acquired Pneumonia
9/4/92	Bulk Tazobactam Amendment
10/8/92	Environmental Assessment Update
11/23/92	First Safety Update
1/28/93	Bulk Tazobactam Amendment - Process Modification
5/21/93	Environmental Assessment Appendices
5/25/93	Second Safety Update
6/15/93	Demographic Subset Analyses
9/7/93	Comments from Cyanamid in responding to FDA draft labeling, received from FDA on 8/13/93.
10/22/93	NDA 50-684 approval received.

ATTACHMENT 5

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

RE: U.S. Patent No. 4,562,073

ISSUED: December 31, 1985

TO: Ronald G. Micetich, et al

FOR: PENICILLIN DERIVATIVES

Honorable Commissioner of Patents and Trademarks
Box Patent Extension
Washington, D.C. 20231

POWER OF ATTORNEY

Taiho Pharmaceutical Company Limited, owner of the above identified United States Patent No. 4,562,073 issued December 31, 1985, hereby appoints Robert B. Murray, Reg. NO. 22,980, of the firm Nikaido, Marmelstein, Murray & Oram, as their attorney to represent them in proceedings in the United States Patent and Trademark Office and the Food and Drug Administration to extend the term of Patnet No. 4,562,073 pursuant to the provisions of 35 U.S.C. Section 156 and to transact all business in the United States Patent and Trademark Office in connection therewith. This Power of Attorney is effective immediately and shall continue unless revoked, until the completion of the indicated proceeding.

Please direct all correspondence to:

Robert B. Murray
Nikaido, Marmelstein, Murray & Oram
Metropolitan Square
Suite 330 - G Street Lobby
655 15th Street, N.W.
Washington, D.C. 20005-5701

Taiho Pharmaceutical Company Limited

Date November 15, 1993

By Yukio Kobayashi

Typed Name Yukio KOBAYASHI

Title President